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Elevated exhaled nitric oxide in anaphylaxis with respiratory symptoms

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FeNO, fractional exhaled nitric oxide;
iNOS, inducible nitric oxide synthase

ABSTRACT

Background: Anaphylaxis is a serious type I allergic reaction that occurs suddenly and can result in death, but it is sometimes difficult to differentiate from other diseases, and physicians must rely on symptoms alone for its diagnosis. Meanwhile, fractional exhaled nitric oxide (FeNO) concentration, used in assessing airway inflammation in bronchial asthma, is known to be affected by atopic disposition. The possible role of FeNO measurements was evaluated in patients with anaphylaxis.
Methods: FeNO was measured in 52 adult patients (17–78 years old, median age 41.5 years) in whom anaphylaxis occurred. These measurements were made within 24 h after onset and after about one month when the patients were symptom-free. In some of these patients, FeNO was measured a third time, two months or more after onset.
Results: The FeNO level in the 52 patients was not significantly different in measurement made within 24 h of onset of anaphylaxis and after one month. However, excluding 9 patients who also had asthma history, the FeNO level in the remaining 43 patients decreased significantly from within 24 h of onset (36.7 ± 27.5 ppb) to one month later (28.8 ± 19.5 ppb). Of these 43 patients, this phenomenon was evident in a group that had respiratory symptoms (31 patients), but it was not seen in a group that did not have respiratory symptoms (12 patients).
Conclusions: Elevation of FeNO was related to respiratory symptoms observed in anaphylactic patients without asthma. Although the mechanism of increased FeNO level is unclear, its usefulness for diagnosis of anaphylaxis must be examined in prospective studies.

Introduction

Anaphylaxis is a severe allergic reaction that occurs rapidly and can lead to death, and early diagnosis and treatment affect outcomes. However, anaphylaxis is sometimes difficult to differentiate from an asthmatic attack, fainting, anxiety/panic disorder, acute urticaria, and other conditions, and there are no diagnostic criteria about which there is consensus. In clinical practice, clinical diagnostic criteria based on medical interview responses and characteristic symptoms established by the World Allergy Organization (WAO) are used.[1] However, excluding cases for which there is an obvious time relationship between symptoms and exposure to a causative agent, such as a bee sting or a specific immunotherapy, or when anaphylaxis occurs after exposure to a known allergen for that patient, physicians must currently rely on symptoms alone for diagnosis of anaphylaxis in situations when this information cannot be obtained.

Fractional exhaled nitric oxide (FeNO) concentration, used in assessing airway inflammation in bronchial asthma, is known to be affected by atopic disposition, rhinitis, smoking, and other conditions.[2–4] Focusing both on the fact that atopic disposition affects FeNO production and that the basic pathology of anaphylaxis is an allergic reaction (immediate allergy) via IgE antibodies, the possible utility of FeNO measurements in anaphylaxis was investigated in this study.

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Table 1
Charcteristics of patients with anaphylaxis whose FeNO were measured both in acute phase and stable phase.

Case	Gender	Age	Allergen	Asthma history	Current treatment for asthma	Current smoking	Serious symptom			Treatment for anaphylaxis		FeNO (ppb)	
							Lower airway symptom	Hypotension	Grade[†]	Adrenaline	Steroid	Acute phase	Stable phase
1	F	50	WDEIAn[‡]	−	−	−	+	−	Moderate	−	−	17	19
2	M	62	WDEIAn	−	−	−	−	−	Severe	−	+	24	31
3	F	29	Oral mite[§]	−	−	+	+	−	Moderate	−	−	36	32
4	M	65	Venom	−	−	−	+	+	Severe	+	+	34	54
5	F	31	Soybean or nuts	−	−	−	+	−	Moderate	−	−	16	12
6	F	19	Unknown drug	−	−	−	+	+	Severe	−	+	10	11
7	M	47	Wheat	−	−	−	+	−	Moderate	−	−	63	35
8	M	25	Anisakis	−	−	+	+	−	Moderate	−	+	20	16
9	M	45	NSAID[¶]	−	−	+	+	−	Moderate	+	+	108	43
10	F	34	Nuts	−	−	−	−	−	Mild	−	−	23	18
11	F	63	Shrimp	−	−	−	−	+	Severe	+	+	28	36
12	F	49	Wheat	−	−	−	+	−	Severe	+	+	29	46
13	M	39	Butterbur sprout	−	−	+	+	−	Moderate	+	+	45	22
14	M	58	Bamboo shoot	−	−	−	−	−	Moderate	−	+	35	35
15	M	31	Unknown	−	−	+	+	−	Moderate	−	−	25	29
16	M	45	Anisakis	−	−	−	+	−	Moderate	+	+	20	15
17	F	68	Anisakis	−	−	−	−	+	Severe	−	+	11	11
18	M	40	Egg	−	−	−	+	−	Moderate	+	+	82	49
19	F	36	NSAID	−	−	+	+	−	Moderate	+	+	14	9
20	F	33	Anisakis	−	−	+	−	+	Severe	+	+	14	8
21	F	20	Wheat	−	−	−	+	+	Severe	−	+	21	14
22	F	47	Anisakis	−	−	−	+	−	Moderate	+	+	45	21
23	M	49	NSAID	−	−	−	+	−	Moderate	+	+	56	59
24	F	57	Unknown	−	−	−	−	−	Mild	−	+	17	13
25	F	73	Mushroom	−	−	−	+	−	Moderate	+	+	21	14
26	M	17	Shrimp	−	−	−	+	−	Moderate	+	−	71	69
27	F	54	NSAID	−	−	+	+	−	Moderate	−	+	37	28
28	M	53	Shrimp	−	−	−	−	−	Moderate	−	+	42	32
29	F	41	Buchwheat	−	−	−	−	−	Moderate	+	+	13	17
30	M	33	Apple	−	−	+	+	−	Moderate	+	+	28	22
31	M	37	Soybean	−	−	−	+	−	Moderate	+	+	82	66
32	F	47	Mackerel	−	−	−	+	−	Moderate	−	+	44	23
33	F	25	Unknown	−	−	−	−	−	Moderate	−	−	21	13
34	F	42	Antibiotics	−	−	−	+	−	Moderate	+	+	137	35
35	F	25	Sea urchin	−	−	−	+	−	Severe	+	−	13	16
36	F	55	Shrimp	−	−	−	+	−	Moderate	+	+	37	50
37	M	54	Spirinchus lanceolatus	−	−	−	+	+	Severe	+	+	5	5
38	F	24	Wheat	−	−	−	+	−	Moderate	−	−	63	22.4
39	F	38	Unknown	−	−	−	+	−	Moderate	+	+	16	15
40	M	61	Antibiotics	−	−	−	+	−	Moderate	+	−	26	25
41	F	30	Shrimp	−	−	−	−	−	Mild	+	+	53	101
42	M	21	Nuts	−	−	−	−	−	Mild	+	+	19	17
43	M	42	WDEIAn	−	−	−	+	+	Severe	+	+	58	28
44	F	52	Anisakis	+	FP[‖](200) 4puff	−	+	−	Moderate	−	−	49	52
45	M	43	NSAID	+	FP(250)/SM[#] (25) 2puff	−	+	−	Moderate	−	+	25	34
46	M	41	WDEIAn	+	No	+	+	+	Severe	−	+	15	26
47	F	47	Shrimp	+	BUD(160)/FM[††](4.5) 2puff	−	−	+	Severe	+	+	23	22
48	F	21	Spawn	+	No	+	−	+	Severe	+	+	42	117
49	F	36	Tuna	+	No	+	+	−	Moderate	+	+	30	14
50	F	34	Supplement drug	+	No	+	−	+	Severe	+	+	31	34
51	F	78	NSAID	+	BDP[‡‡](100) 2puff	−	+	+	Severe	−	+	37	26
52	M	17	WDEIAn or tomato	+	no	−	+	+	Severe	+	+	115	183

[†] Based on reference 6.
[‡] Wheat dependent exercise induced anaphylaxis.
[§] Mite-contaminated foods.
[¶] Non-steroidal anti-inflammatory drug.
[‖] Fluticasone propionate.
[#] Salmeterol.
[††] Formoterol.
[‡‡] Beclomethasone.

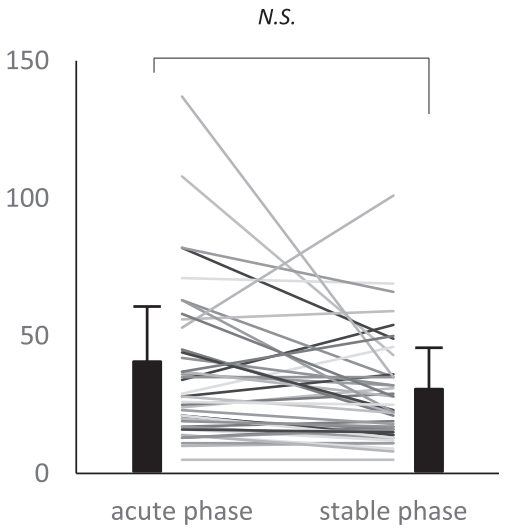

Fig. 1. FeNO levels in 42 patients of anaphylaxis without asthma history at acute phase and stable phase. Bars show mean ± SD.

Methods

Subjects

The investigation was done with 52 adult anaphylaxis patients being diagnosed by World Allergy Organization anaphylaxis guidelines[5] treated at the Allergy Center or Emergency Medical Care Center at the Yokohama City Minato Red Cross Hospital in whom FeNO levels could be measured within 24 h of onset of anaphylaxis and after about one month of onset when patients had no symptoms. Characteristics of all participants of this study was shown in Table 1. The patients' mean age was 42.0 years (17–78 years, median age 41.5 years), and they had a male-female ratio of 11:15. Nine of the 52 patients (15.4%) also had bronchial asthma. The severity of the anaphylaxis[6] was mild, with skin and mucosal symptoms, in 3 patients (5.8%), moderate, presenting with respiratory, circulatory, and gastrointestinal symptoms, in 32 patients (61.5%), and severe, with cyanosis, impaired consciousness, incontinence, and other conditions, in 17 patients (32.7%). FeNO could be measured a third time, two months or more after onset of anaphylaxis, in some patients.

Measurement of FeNO levels

FeNO levels were measured with an expiratory flow rate of 50 mL/s using a NIOX-MINO (Aerocrine AB, Solna, Sweden). Measurements within 24 h of onset were made after the anaphylactic symptoms had improved with treatment. The second measurement was, as a general rule, made when the patients visited the hospital again for a detailed examination of the cause of the anaphylactic symptom episode, about one month after it occurred.

Statistical analysis

All the data were expressed by mean ± SD. Statistical analysis was done with SPSS ver. 21 (IBM Japan, Tokyo), with $p < 0.05$ taken to be a significant difference. Investigation of two paired groups was done with Wilcoxon's signed rank test.

Ethics

This study was approved by the Ethics Committee of Yokohama City Minato Red Cross Hospital (IRB approval number: 2014-27). FeNO samples were collected with patients' informed consent.

Results

The FeNO level in all 52 patients showed a tendency to decrease, from 37.4 ± 27.6 ppb within 24 h after the onset of anaphylaxis to 33.5 ± 30.4 ppb after one month, but the difference was not significant. Since complication of bronchial asthma can affect FeNO level, 43 cases without asthma history were compared with 9 cases with asthma history. As shown in Fig. 1, the FeNO level decreased significantly in these 43 cases from within 24 h after onset (36.7 ± 27.5 ppb) to one month later (28.8 ± 19.5 ppb) ($p = 0.008$). In the 9 cases with asthma history, the FeNO level was 40.8 ± 29.7 ppb within 24 h of onset and 56.4 ± 56.5 ppb after one month, and it showed no significant changes. Then, all analysis of afterward was carried out about the 43 cases without asthma history. In the 43 cases, the FeNO levels in 33 cases whose FeNO was measured a third time two months or more after the onset of

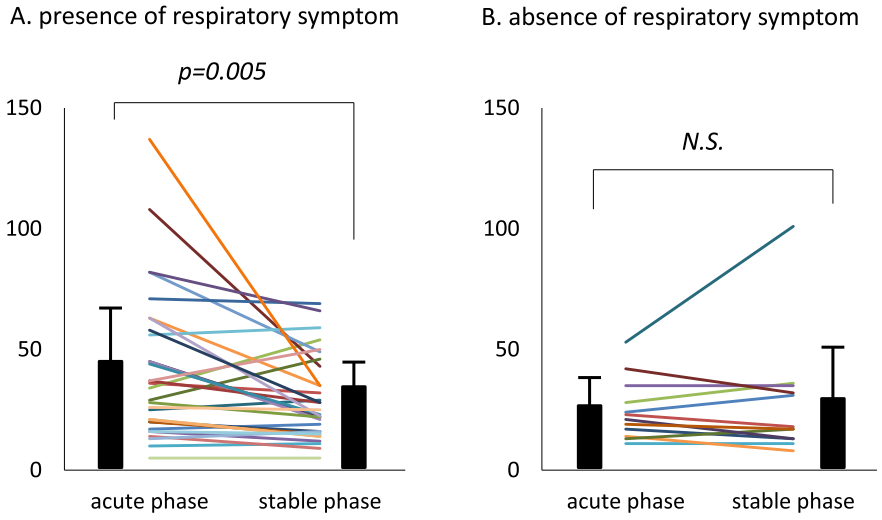

Fig. 2. A. FeNO levels in 30 patients of anaphylaxis associated with lower airway symptoms but without asthma history at acute phase and stable phase. Bars show mean ± SD. B. FeNO levels in 12 patients of anaphylaxis without any lower airway symptoms or asthma history at acute phase and stable phase. Bars show mean ± SD.

anaphylaxis were 35.1 ± 28.7 ppb within 24 h of onset, 25.6 ± 14.5 ppb after one month, and 23.5 ± 14.4 ppb after two months or more. There was no significant difference between the FeNO levels at one month and two months or more after onset. In an analysis by level of severity in the 43 cases without asthma history, a significant decrease was seen in FeNO the onset period of 24 h and after one month (mean 28.4 days after onset) only in moderate cases ($n = 28$). No significant changes were seen in the severe cases ($n = 11$), and the number of mild cases ($n = 4$) was too small for analysis. Focusing on respiratory symptoms, which were the main symptoms in the moderate patients, those with and without respiratory symptoms were compared, and it was found that, in the group with respiratory symptoms ($n = 31$), FeNO levels decreased significantly from 41.3 ± 30.4 ppb within 24 h of onset to 29.2 ± 17.3 ppb after one month ($p = 0.005$) (Fig. 2A), whereas in the group without respiratory symptoms ($n = 12$), no significant change was seen in the FeNO level, which was 25.0 ± 12.7 ppb within 24 h of onset and 27.7 ± 25.1 ppb after one month (Fig. 2B). In addition, FeNO levels at acute phase in comparison with stable phase was significantly high in the cases treated with systemic corticosteroid ($p = 0.042$, $n = 32$) but was not in the cases with adrenaline injection ($n = 25$), suggesting that systemic corticosteroid and adrenaline may affect FeNO in a different way. In concern of current smoking, elevated FeNO level was seen even in the cases with current smokers.

Discussion

Anaphylaxis needs to be differentiated from various diseases for its diagnosis.[5] Peripheral blood tryptase and histamine, which reflect the pathology of a patient, are measured for the purpose of diagnosing anaphylaxis, but conditions in which obvious elevations in these substances are seen are anaphylaxis from bee stings or drugs and serious conditions in which decreased blood pressure occurs. In anaphylaxis due to food allergies or in conditions that do not result in decreased blood pressure, obvious elevations in tryptase and histamine are not seen, so that changes need to be judged over time.[6] In other words, these measurements are useful in severe anaphylaxis in which diagnosis is easy, but their usefulness declines in patients whose diagnosis is unclear, and sensitivity can be considered a major problem. Thus, the diagnosis of anaphylaxis must still depend on symptoms, and the development of a simple diagnostic marker is awaited.

In this study, it was found that the FeNO level measured within 24 h of onset in anaphylaxis patients, excluding those with concurrent bronchial asthma, was significantly higher than levels during the stable phase after one month or more had passed from the acute phase, after which a constant level was maintained. This result strongly suggests that, although FeNO levels at normal times before the onset of anaphylaxis are unknown, the change in the FeNO level is related to the pathology of anaphylaxis. As mentioned above, there are currently no pathological markers for which results can be obtained simply and in a short time and that can be used in clinical practice. It is impossible to examine the sensitivity and specificity of FeNO for diagnosis of anaphylaxis, because we had not measured FeNO with the control cases who visited us as suspicious anaphylaxis but were diagnosed as other diseases. Nonetheless, it is possible that measurement of FeNO levels may become a surrogate marker of diagnosing anaphylaxis accompanied with lower respiratory symptoms. That these changes are seen, and that the FeNO level is elevated in the acute phase of anaphylaxis in patients with respiratory symptoms alone, is a plausible result considering that the airway is the source of FeNO.

Nitric oxide (NO) is known to be produced from airway epithelial cells and macrophages. It is produced physiologically even in healthy people, but in bronchial asthma, the expression of inducible nitric oxide synthase (iNOS) from stimulation of inflammatory cytokines and other substances increases, large amounts of NO are produced, and respiratory levels rise.[7] In addition to airway epithelial cells and macrophages, NOS is expressed in vascular endothelial cells, platelets, mast cells, renal epithelium, and the hippocampus in the brain. Induced NO plays important roles in various diseases, but in anaphylaxis, it is thought to contribute to making the pathology more severe via vasodilation.[8] Cauwels et al.,[9] using a murine model of anaphylaxis induced by platelet activating factor (PAF), reported that NO plays a central role in the pathology of anaphylaxis since NG-nitro-L-arginine methyl ester (L-NAME), a NOS inhibitor, completely prevented the death of the mice. Since anaphylaxis is a systemic allergic reaction, there is a sufficient likelihood that expression of iNOS is increased in airway epithelium together with vascular endothelium, and expression of iNOS draws attention as the causes of the rise in FeNO in the acute phase of anaphylaxis shown in this study. Sade et al.[10] analyzed the expression of iNOS and endothelial nitric oxide synthase (eNOS) in a mouse model of anaphylaxis using PCR and immunohistochemistry. Results was a significant increase in iNOS mRNA expression and NO production as early as 10–30 min after allergen challenge in both heart and lungs, while the expression of eNOS mRNA was not altered during the course of the experiment, indicating an involvement of iNOS in the immediate physiological response of anaphylaxis.

In concern of elevated FeNO in this study, there remains the possibility of subclinical/undiagnosed asthma even if asthma histories were denied by interview. We measured airway hyperresponsiveness of only two participants of the study (case 43 and 52 in Table 1). One patient (case 43) without asthma history showed FeNO elevation at acute phase of anaphylaxis, and Astograph® being performed two months after anaphylactic episode revealed no airway hyper-responsiveness. Another severe anaphylactic patient (case 52) with asthma history in his infant period, whose FeNO level was high but did not alter during the following period, showed a potent airway hyper-responsiveness. Thus, the presence of subclinical/undiagnosed asthma must be ascertained by the test of airway hyper-responsiveness in all anaphylactic patients to examine the pathogenesis of elevated FeNO levels in acute phase of anaphylaxis in future studies.

A definitive diagnosis of anaphylaxis is important not only in the acute phase but also in relation to preventing subsequent recurrence. Despite later detailed examinations of causes, the allergen cannot be identified in a certain percentage of anaphylaxis patients. In these patients, the course is observed without ever knowing for certain whether the acute phase episode was actually anaphylaxis. The FeNO measurements described herein can be easily performed even during anaphylaxis except in patients with impaired consciousness. In conclusion, although the mechanism of increased level of FeNO is unclear and its role in pathogenesis is unknown, its usefulness for diagnosis of anaphylaxis must be examined in prospective studies.

Conflict of interest

The authors have no conflict of interest to declare.

Authors' contributions

YN designed the study and wrote the manuscript. YH, JE, MF and KY contributed to data collection. AI performed the statistical analysis and interpretation of the results. All authors read and approved the final manuscript.

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